

Figure 1

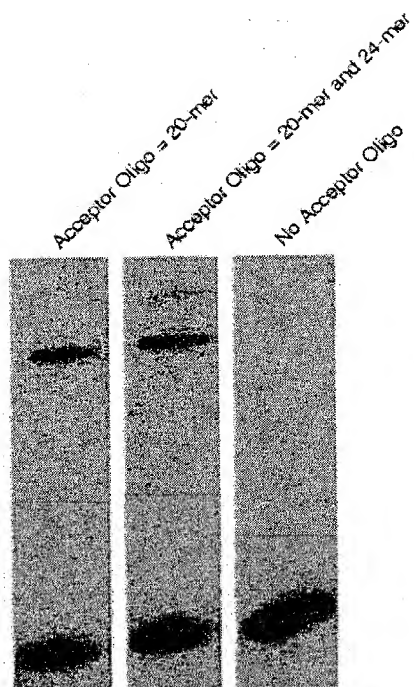


Figure 2

OPS at Topol cleavage site

↓

TCRIBamS: 5' AATTCGCGGCCGCAAAAAGACTT--GATC 3'

TCRIBamAS: 3' GCGCCGGCGTTTTTCTGAA--CTAG--DMT 5'

Figure 3A

OPS at Topol cleavage site

↓

TCRBstBS: 5' AATTCGCGGCCGCAAAAAGACTT--CG 3'

TCRBstBAS: 3' GCGCCGGCGTTTTTCTGAA--GC--DMT 5'

Figure 3B

TCRIBamS: 5' AATTCGCGGCCGCAAAAAGACTTGATC 3'

TCRIBamAS: 3' GCGCCGGCGTTTTTCTGAACTAG 5'

Figure 3C

GATC removed on topo cleavage to give BamHI compatible 5' overhang

5' AATTCGCGGCCGCAAAAAGACTTGATCAAGTCTTTTTGCGGCCGCG 5'

3' GCGCCGGCGTTTTTCTGAACTAGTTCAGAAAAACGCCGGCGCTTAA 3'

Figure 3D

Figure 3
A - D

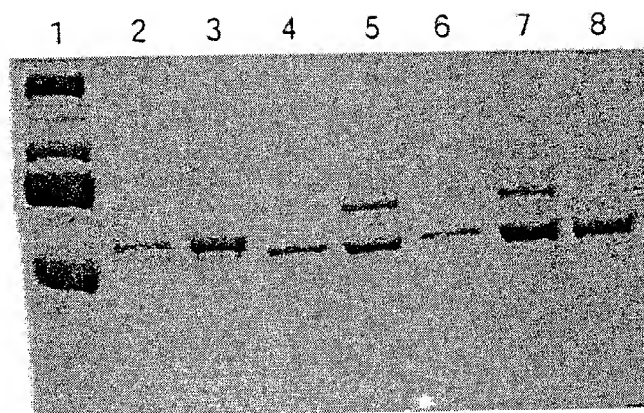


Figure 4

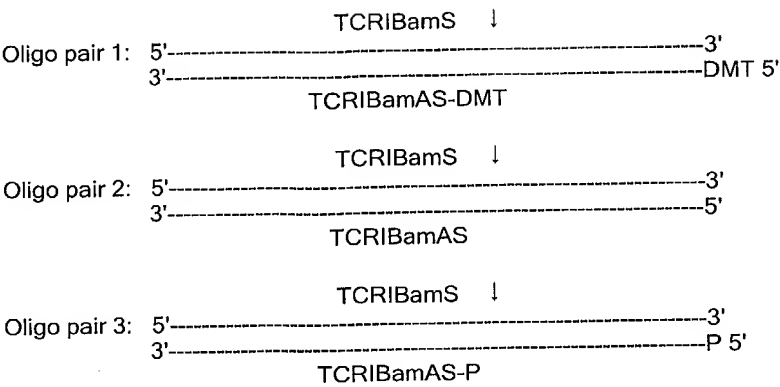


Figure 5A

“↓” indicates site of topo cleavage

Oligo pair 4: TCR1BstBS + TCRABstBAS-DMT
Oligo pair 5: TCR1BstBS + TCRABstBAS
Oligo pair 6: TCR1BstBS + TCRABstBAS-P

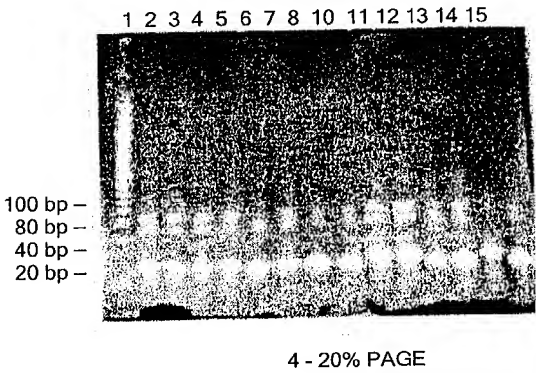


Figure 5B

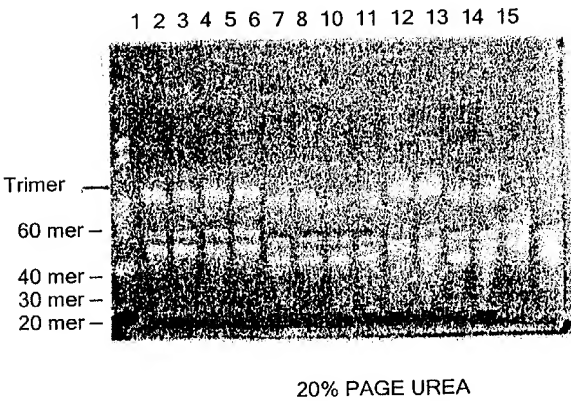


Figure 5C

Figure 5
A - C

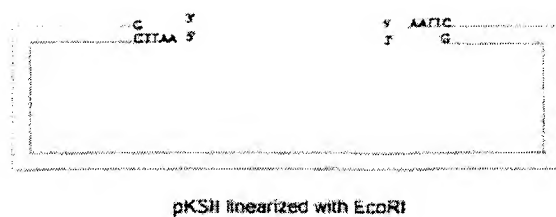
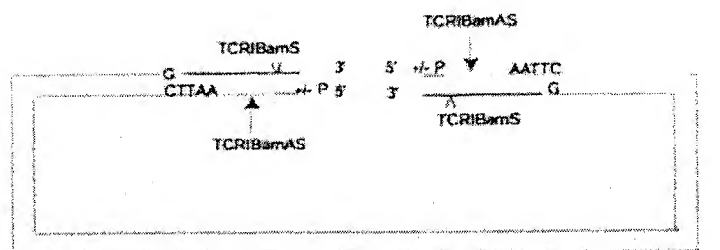


Figure 6A



GATC overhang left after Topo cleavage and removal of 4bp downstream from Topo cleavage site

site of Topo cleavage

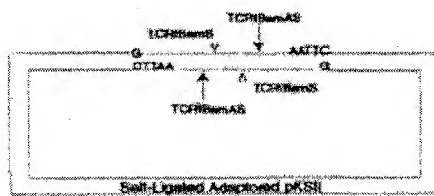


Figure 6B

Sequence is:
AATTGCGGCGCG

Figure 6C



Figure 6D

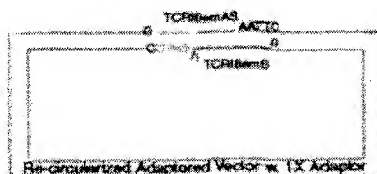


Figure 6
A - D

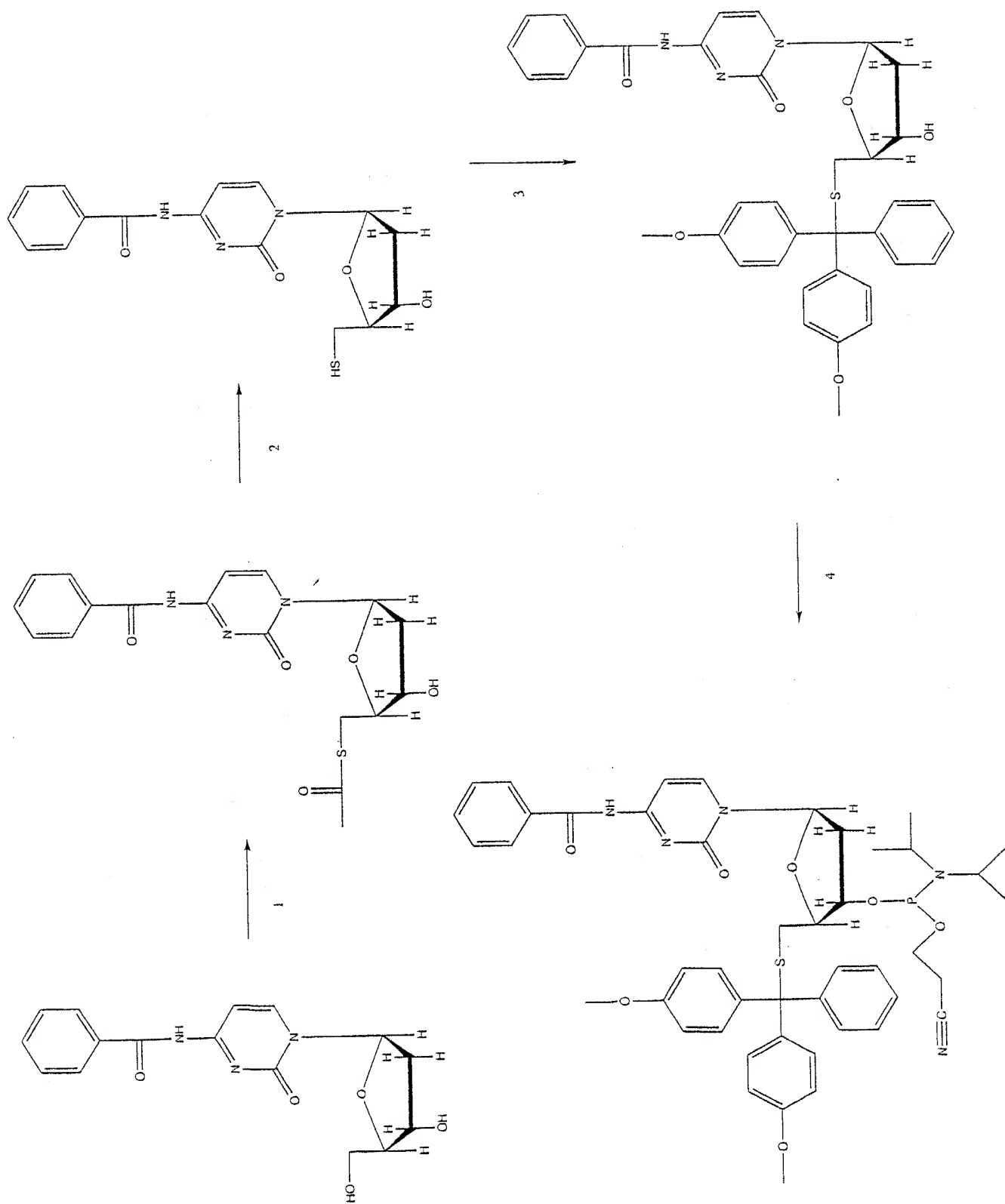
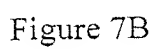


Figure 7A



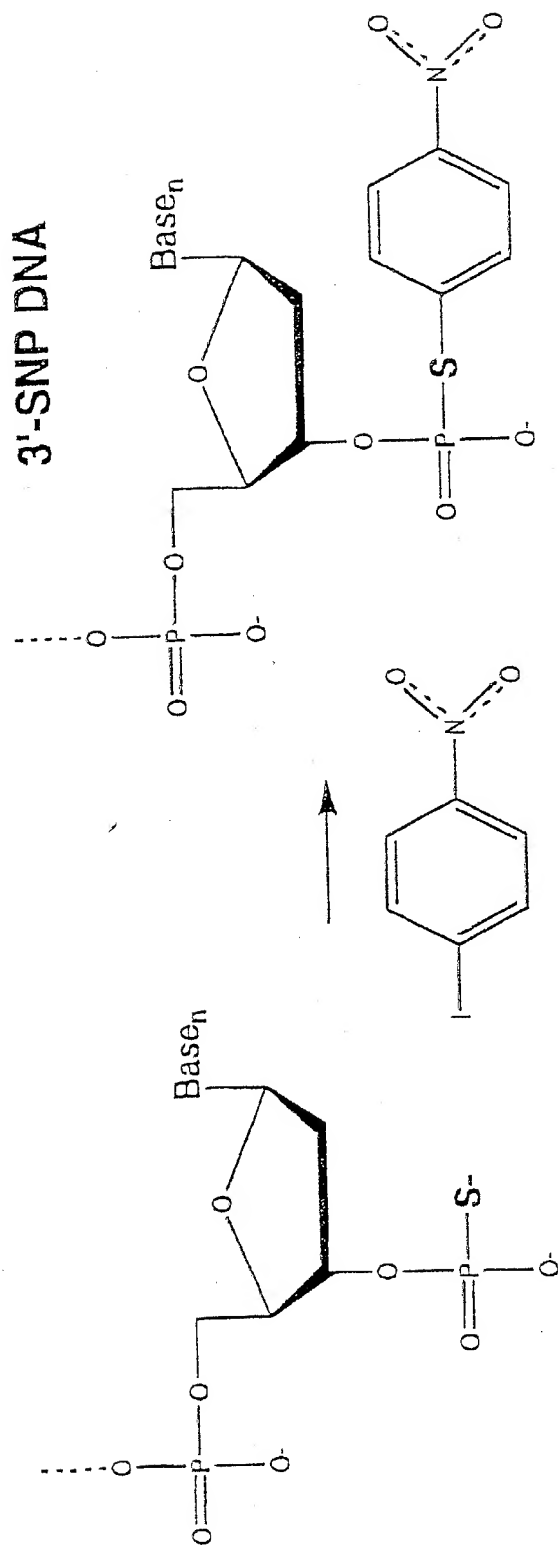


Figure 8